The lily disease in Bermuda.

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(WITH PLATE I.)

In the winter of 1888, through the kindness of General Russell Hastings, of Hamilton, Bermuda, I had the opportunity of investigating a disease, epidemic in the lily fields there from March to July. Diseased specimens had previously been shown to several naturalists who had visited Bermuda, and a few of the same had been sent to the United States for examination. None of those who saw these specimens were able, upon a merely cursory examination, to identify the disease; so that the subject still remained open for investigation.

Towards the end of February I sailed for Bermuda, in order to observe personally the phenomena attendant upon the first stages of the disease, which usually begins early in March. A few words on the cultivation of the lily in Bermuda and upon the history of the disease may not be out of place.

The industry is a comparatively new but prosperous one, being only a few years old. The lily cultivated is the so-called Lilium Harrisii, a dwarf variety of Lilium longiflorum. This variety has been propagated from year to year by bulb-scales and by a few plants raised yearly from seed. The latter are always more vigorous than those raised from bulb scales; they tend, however, to return to the original longiflorum type. The bulbs, planted in the early autumn, grow throughout the winter months and flower in the spring. They are taken up again early in the summer, and shipped to the United States, where a good market is also found for the flowers in the spring months. The lilies are planted in rows, in small fields; these fields are surrounded by high oleander hedges, which serve to protect the plants from the wind.

So far as I know, the lily disease was first noticed about 1885, since which time it has yearly become more serious, until at present it threatens to do serious damage to the lily crop. It makes its appearance in the spring when the days begin to grow warm. This marked difference in the temperature between day and night results in a heavy fall of dew, so that even after the sun is high in the heavens

large dew drops may be found upon the lily leaves. A little cool weather, or a few warm, dry days free from dew, will check the activity of the disease, while a warm, damp day will cause it to spread with great rapidity. The disease first appears as a minute orange-colored spot upon a leaf or flower, usually on the upper side of the leaf. Fig. 1 (*a*) shows an early stage. The spot gradually increases in size, and finally spreads throughout the whole leaf. In this way whole plants may be killed, and only the stalks left standing. It is not unusual to find several of these spots on the same leaf, as in fig. 1. If, for any reason, the progress of the disease is checked, these become dry, leathery patches of a buff color.

It was thought possible, by some of those who had seen the diseased specimens, that the spots might be due to the stings of insects. Though at first this did not seem unlikely, a closer acquaintance with the disease rendered it highly improbable.

Sections of the spots examined under the microscope revealed nothing more than that in the diseased area the structure of the leaf had collapsed; while along the edges of the spot the cells were somewhat swollen and the cell walls thickened. The cell contents were slightly shrunken, and contained a number of bright dots which did not stain with the ordinary reagents. In a few cases where specimens were teased an occasional hypha of a fungus was found, which, however, might easily have come from external contamination. In a slightly more advanced stage of the disease hyphæ of a fungus might be found ramifying through the soft decaying tissues, the gonidiophores appearing on the surface (fig. 2).

This fungus invariably appears in advanced stages of the disease; moreover, it is always the first to appear. Other fungi, such as Macrosporium, Eurotium, and Penicillium, also appear on the decaying leaves, but this fungus invariably precedes all others. At the same time, as it could never be found until the tissue had become pretty thoroughly rotten, it seemed at first sight to be rather a consequence than a cause of the disease.

Early in April I was obliged to return to the United States, and for a time my investigations were interrupted to be resumed again in the summer, when I had specimens forwarded to me from Bermuda. It had seemed possible that the disease might be due to bacteria, on account of its pecul-

iarly rapid growth and its susceptibility to climatic conditions, as well as for other reasons. Accordingly a series of cultures in nutrient gelatine and agar-agar was begun. By this method two distinct species of bacteria were isolated from the spots, either of which might have been the cause of the disease.

Inoculations were attempted, both in the laboratory and in the garden, all of which failed to produce the disease, but as the American climate is dry, and therefore unfavorable, it was thought that the bacteria had not been given a sufficient trial, and so it was decided to let the whole matter lie over until the next winter, when inoculation experiments could be made in Bermuda.

Shortly before visiting Bermuda for the second time my attention was again directed to the fungus as a cause of the disease by the appearance of Professor Marshall Ward's description of a similar disease caused by a fungus growing upon the Lilium candidum in England. Upon my arrival in Bermuda, in February, 1889, I renewed my efforts at inoculation with bacteria, but in every case without success. I also placed a number of plants in glass jars which contained fluid cultures, to see if by any chance the action of bacteria upon the bulb might not have something to do with the epidemic. These plants, however, remained remarkably healthy, as did also a number of plants from which I removed the bulbs, placing the stalks in fluid cultures, so that the bacteria had direct access to the tissues of the plants. The weather was extremely unfavorable, so that although I saw numerous small spots, I met with none large enough to show gonidiophores of the fungus. I was, therefore, unable to complete my experiments satisfactorily in Bermuda, as I could not even get a culture of the fungus as a starting point. Since my return, however, I have had specimens sent to me and have been able to carry on a series of experiments in the laboratory, by means of which I have, at length, ascertained definitely the cause of the disease. Meanwhile I have repeatedly tried to inoculate plants in the laboratory with bacteria, all of which attempts have been signal failures, as have been likewise all attempts to produce the disease by fungus spores sown in a drop of Irish moss which had been placed upon a leaf. In the latter case it was found impossible (without resort to artificial means) to keep the air of the room moist enough so that the drop of Irish moss would not dry up soon after it was put upon the leaf. I readily succeeded, how-

ever, in getting numerous Irish moss cultures of the fungus, and I have also grown it in water both ordinary and sterilized, and in hanging drops as well as in drops merely placed upon a slide.

In my next experiments leaves, freshly picked from the plant, were used and kept fresh in a beaker of water, which was placed under a bell glass, where there was a second beaker of steaming water, which was occasionally renewed so that the atmosphere was kept warm and moist, thus making the conditions highly favorable. Bacteriological experiments under these conditions failed as in all previous experiments. On the other hand, experiments with the fungus were entirely successful. If a drop of Irish moss, placed on the surface of a leaf is inoculated with spores of the fungus mentioned above, a disease spot will appear in about two days, unless in some way the drop has dried up, or been otherwise disturbed. If the drop has simply dried up, it is only necessary to moisten it again, and the spot will soon appear. About the third or fourth day the gonidiophores appear, and soon bear compact bunches of gonidia, the whole forming a downy growth on both sides of the leaf. It makes no difference whether the original spores are sown in a drop on the upper or the under side of the leaf. In either case the disease is produced. I have substituted drops of water for the Irish moss as a culture medium with equal success, and to exclude contamination, drops of sterilized water were used with which I also obtained excellent results.

To prove beyond all doubt that the fungus is the cause of the disease, the following experiment was made: Into a tube of sterilized nutrient gelatine some fungus spores were introduced on the tip of a sterilized needle. The contents of the tube were then "plated" according to Koch's method. The fungus grows luxuriantly in this medium, and can thus be obtained free from bacteria or other fungi. Spores are less quickly produced on the rich gelatine than on the comparatively poor Irish moss. Inoculations made with spores from a pure culture of this kind readily produced the characteristic spots, while blank control experiments showed nothing whatever.

The fungus appeared to me to be identical with the Botrytis recently described by Prof. Marshall Ward[1] as growing upon the Lilium candidum in England, and from a specimen sent to him Professor Ward has identified it as the same.

[1] A Lily Disease, by H. Marshall Ward, M. A., F. R. S., F. L. S. Annals of Botany, vol. ii. no. 7, Nov., 1888.

So far as I know the Lilium candidum is exempt from the disease in Bermuda, but this species is only occasionally grown there, and then with only a few together. Under similar circumstances the longiflorum also is generally exempt, the plants in the fields alone suffering. The true longiflorum is not considered to be as susceptible to the disease as the Harrisii. Since Marshall Ward has described the Botrytis and its method of growth in so complete a manner, it would be superfluous for me to enter upon a detailed description of it. I shall, therefore, give but a brief summary of the main facts in its life history, as I have observed them.

The gonidia are ovoid in shape, about 0.02 mm. long and about 0.015 mm. broad. They are, at first, colorless, but when mature are light brown, the whole gonidiophore passing through the same change of color. The gonidia are attached to the gonidiophores by means of slender sterigmata. These sterigmata are frequently found still adhering to the spore after it is entirely separated from the gonidiophore upon which it grew. While the normal shape of the spore is ovoid, spores are often found of different shapes, as those depicted in fig. 7. These forms, however, seem to belong to the period of germination, being simply conditions which the gonidia assume in the process of swelling. The spores are almost always ovoid, although in some old bunches they may sometimes be elongated and even divided by a partition. In germination the gonidium swells, the protoplasm becoming quite dense. Soon after, the wall begins to bulge out in several places, from which hyphæ are eventually protruded. One of these is generally more vigorous than the others, and grows much more rapidly, producing a complicated mycelium, while the other hyphæ are scarcely more than buds. The protoplasm of the rapidly growing tips of the hyphæ is quite dense, while that in the older hyphæ is clear and much vacuolated. Two remarkable features of the mycelium described by Marshall Ward I have, also, observed. These are, first, organs of attachment, consisting of thick cone-shaped tufts (fig. 5), developed upon the hyphæ when they come in contact with any foreign body which they are not able to penetrate. The second peculiarity is the development of cross branches between contiguous hyphæ, thus forming a network in the mycelium (fig. 6). The gonidiophores are, on the leaves, as a rule, about one mm. in height. When they reach this height, their tips begin to swell, and numerous small peg-like processes appear upon the heads

thus formed. These are the sterigmata, and on each of these a gonidium is formed (fig. 3). There are seldom fewer than four gonidia in a head, and they usually bear a much larger number. With perhaps thirty of these heads to the sq. mm., which is about the average number on an ordinary diseased leaf, it is not strange that, under favorable conditions, the disease should spread from leaf to leaf and from plant to plant, with surprising rapidity. If the growth of the fungus is unchecked, the erect hyphæ may bud out just below the gonidiophores, and after growing a short distance produce another bunch of gonidia, and so on, thus forming a series such as is shown in fig. 4.

In this disease the fungus does not grow in the sound tissues of the host, extracting nourishment from them, but the spores, germinating upon the surface of the leaf, in some way macerate it. According to Marshall Ward, this is done by means of a ferment secreted by the tips of the hyphæ. I have not as yet been able to experiment upon this ferment, but I have seen nothing to invalidate his view. The cell walls having been softened, the hyphæ pass through them, ramifying amongst the decaying tissues of the leaf.

The principal conclusions arrived at in this paper are the following:

1. A disease, hitherto unexplained, and threatening to become a serious epidemic in the lily fields of Bermuda, has been assigned to a definite cause.

2. There is no evidence that the blight is due to the stings of insects.

3. There is no evidence that the disease is caused by bacteria.

4. There is strong evidence that the disease is caused by a fungus, growing upon and within the leaves or flowers.

5. The fungus which causes the Bermuda lily disease is identical with the Botrytis recently described as the cause of a similar disease in England.

In making the investigation just described it was hoped not only to discover the cause of the disease, but also to suggest remedial measures. In this connection I may remark that the fungus itself seems to be delicate, so that its growth might probably be checked by almost any of the poisons used in the treatment of plant diseases, were it not for the difficulty of applying these effectively. Either a powder sprink-

led on the leaves, or a liquid sprayed upon them would here be ineffective, for the moisture would soon remove them, leaving the fungus free to grow. If, however, the fungus were present at the time of the application of the remedy, that part of it growing outside of the leaf might be killed, although that would not prevent the mycelium within the leaf from continuing to grow.

I have, however, observed that plants growing under the shelter of the oleander hedges are remarkably exempt from the disease. This I attributed at first to the drippings from the leaves, which I thought might partake of the acrid character of the sap of the oleander. I am inclined to believe, however, that it is rather because the lilies under the hedges are remarkably free from moisture. In fact, the overhanging hedge collects most of the dew, so that the dew drops do not readily gather upon the leaves of the lily plants, and thus, perhaps, no opportunity is given for the growth of the fungus.

It might, therefore, be found feasible to grow, in alternate rows with the lilies, some other crop, which, being higher and having more spreading foliage, should keep off the dew. A row of stakes with branches wattled among them might serve the same purpose and check the virulence, even if it did not entirely prevent the recurrence of the disease.

EXPLANATION OF PLATE I.—Fig. 1, Upper side of a leaf, with two spots upon it, showing different stages of the disease; (*a*) an early, and (*b*) a more advanced stage. Fig. 2, A section through a diseased spot when the disease is far advanced. The hyphæ occupy all the tissue in the diseased area. Fig. 3, A gonidiophore with its gonidia, showing the sterigmata and the attachment of the gonidia. Fig. 4, A series of bunches of gonidia on an old gonidiophore. Fig. 5, An organ of attachment. (After Marshall Ward.) Fig. 6, Part of a mycelium showing cross branches. Fig. 7, Gonidia; (*a*) an ordinary gonidium; (*b*) and (*c*) forms frequently assumed by gonidia shortly before germination. Fig. 8, A sprouting gonidium.

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